

# Susceptibility of Fruit from Diverse Apple and Crabapple Germplasm to Attack by Plum Curculio (Coleoptera: Curculionidae)

CLAYTON T. MYERS,<sup>1,2</sup> TRACY C. LESKEY,<sup>1</sup> AND PHILIP L. FORSLINE<sup>3</sup>

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**ABSTRACT** Plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), is an important apple, *Malus domestica* Borkh., pest that significantly hinders sustainable apple production in eastern North America. The potential for host plant resistance to plum curculio among apple germplasm has never been rigorously evaluated. Thus, studies were conducted to assess the susceptibility of a number of exotic and domestic *Malus* accessions housed at the USDA Plant Genetic Resources Unit (PGRU) “core” collection in Geneva, NY. Contrary to earlier published reports and promising data from a field assessment in 2005, these results suggest that there is probably little potential for genetic resistance to plum curculio among the *Malus* germplasm collection evaluated. More specifically, four *Malus* hybrid selections that have previously been released with claims of plum curculio resistance were shown to be susceptible to plum curculio attack. Because there are additional accessions housed at PGRU outside of the core collection that are currently classified as resistant, further studies are necessary to evaluate the true resistance qualities of these releases. It is also important to clarify such discrepancies in both the USDA online Germplasm Resources Information Network and in the horticultural literature. Although other *Malus* species exhibited some variability in fruit susceptibility, none could be classified as being truly resistant to plum curculio attack by any definition that would have relevance to commercial production and sale of apples.

The plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae) is a key pest of apple, *Malus domestica* Borkh., in eastern North America (Racette et al. 1992, Vincent et al. 1999). Adults immigrate from overwintering sites to host fruit trees in the early spring where they mate (Smith and Salkeld 1964, Racette et al. 1992, Piñero et al. 2001). Adults feed on fruit buds and developing fruit, and females oviposit in developing fruit soon after petal fall (LaFleur and Hill 1987, Chouinard et al. 1993) by cutting a small crescent-shaped flap in the fruit skin and then depositing an egg (Quaintance and Jenne 1912, Chapman 1938). Subsequent larval feeding can lead to either fruit drop (Levine and Hall 1977) or severe scarring (Quaintance and Jenne 1912, Racette et al. 1992). Efforts aimed at control of plum curculio have been extensive; most apple growers rely on chemical insecticides for plum curculio management, which presents a challenge to sustainable apple production.

Some past research has been conducted on the breeding of apple cultivars resistant to key apple pests (Goonewardene et al. 1975, 1979; Goonewardene and Kwolek 1985; Plourde et al. 1985; Goonewardene 1987; Goonewardene and Howard 1989). Most of this work was conducted with disease-resistant *Malus* selections

from the Purdue-Rutgers-Illinois (PRI) cooperative breeding program (Crosby et al. 1992), including several selections that were released with claims of multiple pest resistance. However, little to no follow-up research has been done to corroborate these preliminary findings, and widespread industry adoption of these “pest resistant” cultivars has not occurred. Clarification of the true resistance status of these cultivars is necessary to focus future breeding efforts on germplasm sources that exhibit true resistance traits.

Beyond evaluations of PRI cultivars, little work has been published on the pest susceptibility of other *Malus* species, which may contain alleles for insect-resistant traits. Resistance to woolly apple aphid, *Eriosoma lanigerum* (Hausmann) (Knight et al. 1962); rosy apple aphid, *Dysaphis plantaginea* (Passerini) (Alston and Briggs 1970); and *Sappaphis devector* (Walker) (Alston and Briggs 1968, 1977) in apple has been described. In terms of fruit-feeding pests, apple maggot, *Rhagoletis pomonella* (Walsh), has been shown to exhibit differential levels of adult oviposition and larval mortality on certain species of crabapples (Neilson 1967; Pree 1970; Reissig et al. 1990; C.T.M. et al., unpublished data). Besides the aforementioned work with PRI cultivar releases and commercial *M. × domestica* Borkh cultivars, no published information is available regarding the pest susceptibility of exotic *Malus* germplasm to attack from plum curculio.

Research has recently been initiated to examine wild and domestic *Malus* germplasm for resistance to several important fruit-feeding insect pests, including

<sup>1</sup> Appalachian Fruit Research Station, USDA-ARS, 2217 Wiltshire Rd., Kearneysville, WV 25430.

<sup>2</sup> Plant Genetic Resources Unit, USDA-ARS, Cornell University, 630 W. North St., Geneva, NY 14456.

<sup>3</sup> Corresponding author, e-mail: clayton.myers@ars.usda.gov.

**Table 1.** *Malus* accessions observed for field assessments and used in laboratory assays are listed with the corresponding identification (PI) number as used in the USDA Germplasm Resources Information Network<sup>a</sup> (GRIN) database, with prior plum curculio resistance claims, and field locations of trees at the core germplasm collection, Plant Genetic Resources Unit<sup>b</sup> (PGRU–Core), Geneva, NY, and Block #54, Appalachian Fruit Research Station<sup>c</sup> (AFRS), Kearneysville, WV, 2005–2006

| Species                                      | Cultivar   | GRIN PI no. | Plum curculio resist. <sup>?</sup> | PGRU–core block, row-tree | AFRS block #54, row-tree |
|--|------------|-------------|------------------------------------|---------------------------|--------------------------|
| <i>M. × domestica</i> Borkh.                 | Delicious  | 589841      | N                                  | 6-15, 9-51, 10-60         |                          |
| <i>M. × domestica</i> Borkh.                 | Liberty    | 588943      | N                                  | 1-53, 6-1, 8-29, 10-65    |                          |
| <i>M. × domestica</i> Borkh.                 | Emilia     | 123989      | N                                  | 1-29, 5-4, 7-41, 10-37    |                          |
| <i>M.</i> hybrid                             | PRI 1312-6 | 590079      | Y <sup>d</sup>                     | 3-28, 6-49, 8-17, 11-49   |                          |
| <i>M.</i> hybrid                             | PRI 1732-2 | 589946      | Y <sup>d</sup>                     | 3-32, 4-64, 8-30, 12-65   |                          |
| <i>M.</i> hybrid                             | Co-op 15   | 589805      | Y <sup>d</sup>                     | 1-11, 6-37, 8-3, 10-16    |                          |
| <i>M.</i> hybrid                             | E36-7      | 589570      | Y <sup>e</sup>                     | 1-24, 4-57, 8-45          | 2-5                      |
| <i>M.</i> hybrid                             | E11-24     | 589571      | N                                  | 1-20, 5-24                | 2-19, 5-1                |
| <i>M.</i> hybrid                             | E14-32     | 589572      | N                                  | 6-64, 8-49, 12-28         | 2-6, 3-2, 4-3, 5-6       |
| <i>M.</i> hybrid                             | E7-47      | 590069      | N                                  | 1-25, 5-30, 9-6, 11-38    | 2-9                      |
| <i>M.</i> hybrid                             | E7-54      | 590070      | N                                  | 1-26, 4-15, 10-42         | 2-16, 3-10, 4-5, 5-2     |
| <i>M.</i> hybrid                             | E29-56     | 590071      | N                                  | 1-22, 5-65, 7-61, 10-11   | 2-18, 3-5                |
| <i>M.</i> hybrid                             | E31-10     | 590072      | N                                  | 1-23, 6-22, 7-27          | 2-17, 3-9, 4-6           |
| <i>M. × robusta</i> (Carr.) Rehd.            | Korea      | 589003      | N                                  | 3-8, 4-35, 8-6, 10-50     |                          |
| <i>M. fusca</i> (Raf.) Schneid.              |            | 589933      | N                                  | 2-7, 4-41, 9-37           |                          |
| <i>M. sieboldii</i> (Regel) Rehd.            |            | 589749      | N                                  | 2-51, 6-18, 7-6, 12-42    |                          |
| <i>M. bhutanica</i> (W. W. Sm.) J. B. Phipps | Macrocarpa | 588930      | N                                  | 2-64, 6-19, 7-38          |                          |
| <i>M. gunnanensis</i> (French) Schneid.      | Vilmorin   | 271831      | N                                  | 3-14, 5-40, 7-19, 12-49   |                          |

<sup>a</sup> Available online at <http://www.ars-grin.gov/npgs/index.html>.

<sup>b</sup> Plant Genetic Resources Unit, USDA–ARS, Cornell University 630 W. North St., Geneva, NY 14456.

<sup>c</sup> Appalachian Fruit Research Station, USDA–ARS, 2217 Wiltshire Rd., Kearneysville, WV 25430.

<sup>d</sup> USDA 2006.

<sup>e</sup> Goonewardene 1987.

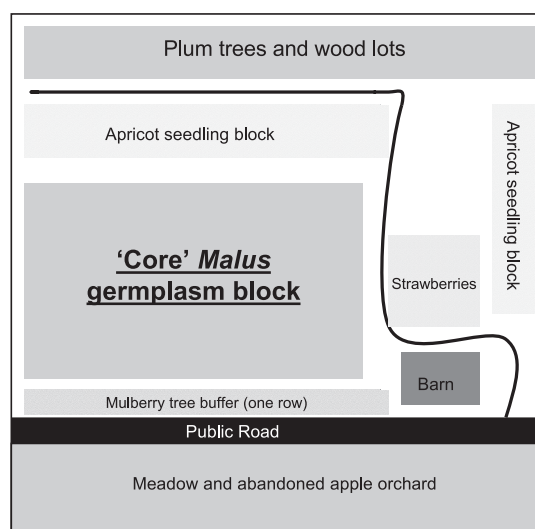
plum curculio. Identification of resistant accessions will help apple breeders to carefully select the most promising germplasm for development of apple cultivars that are naturally resistant to attack from insect pests. This article presents data obtained from laboratory- and field-based studies conducted in 2005–2006, designed to quantify the susceptibility of several *Malus* accessions to direct fruit damage from plum curculio adults and to measure survivorship of plum curculio larvae in fruit.

## Materials and Methods

**Field Assessments, 2005–2006.** Fruit from various *Malus* accessions and hybrid selections were examined for plum curculio adult feeding, oviposition damage, or both via on-tree visual inspection in the field. All accessions used in 2005 and/or 2006 are listed in Table 1. ‘E36-7’ (Goonewardene 1987), ‘PRI 1312-6’, ‘PRI 1732-2’, and ‘Co-op 15’ (USDA 2006) have been reported as resistant to plum curculio. In 2005, one field assessment was conducted at the USDA Plant Genetic Resources Unit (PGRU) “core” *Malus* germplasm collection in Geneva, NY. The core collection (Grauke et al. 1995, Kresovich et al. 1995) for apple includes 206 diverse accessions out of the total collection of 2,438 clonal accessions as described by Forsline (1996). The core collection was planted in 1991 in a replicated block on the Darrow Farm of the New York State Agricultural Experiment Station and consists of the various *Malus* accessions grafted onto M 27 rootstocks. The block is currently situated among a number of apricot, *Prunus armeniaca* L., seedling plantings, a strawberry, *Fragaria ananassa* L., planting, and it is adjacent to a fallow meadow containing an abandoned

apple orchard at its distal end ≈300–400 m away from the core collection. A schematic of this planting and its surrounding landscape is given in Fig. 1.

The management history of the orchard block housing the core collection, from its establishment in 1991 until our studies began in 2005, included a program of fungicides, and insecticides similar to those used in commercial blocks. Removal of insecticides from the orchard management program during the 2005 growing season allowed for the resident plum curculio



**Fig. 1.** Layout map of the core *Malus* germplasm collection block and the surrounding landscape, Geneva, NY, 2005–2006. Map not exactly to scale.

population to increase, resulting in higher pest pressure in the 2006 season. Although there is some variation in tree size due to growth habits of the various accessions, no trees were taller than 4.0 m. Crop load varied somewhat between 2005 and 2006 on some accessions (presumably due to biennial bearing tendencies of some cultivars), but no obvious correlations between accession, crop load, and pest damage were observed among the specific accessions of interest to this study. Although resident plum curculio populations were not assessed via trapping, these populations were assumed to be moderate to high, based upon the past history of the block and preliminary fruit damage observations.

The 2005 PGRU field assessment was conducted on 28 June and over the course of the following week. In total, 70 fruit per tree (with a minimum of 40 fruit per tree if the tree was lightly cropped) from each accession were visually examined; most accessions had at least two replicate trees within the block. In 2006, follow-up field assessments were conducted twice at the PGRU site on selected accessions, once in the mid-season (20 June) and again later in the season (9 August), to account for damage caused by long-living adults and possibly by a second summer generation. In 2006, assessments were made of 100 fruit per tree (with a minimum of 50 per tree if the tree was lightly cropped). Because fewer accessions were evaluated in the field in 2006 than in 2005, the sample size (number of fruit assessed per tree) was increased to enhance the study's statistical rigor.

Field assessments also were conducted in 2006 on various *Malus* hybrid selections planted in block #54 at the USDA–Appalachian Fruit Research Station (AFRS) in Kearneysville, WV. These trees were budded on M7 rootstocks and planted in 1989. Some of the selections were replicated, whereas others were not. This block was surrounded on two sides by woodlots, making it an ideal haven for plum curculio activity. It also was bordered by a block of pear seedlings as well as fallow meadows. A schematic of this planting and its surrounding landscape is given in Fig. 2. Three assessments were conducted at this site: one assessment in the early season (8 June), one assessment in mid-season (13 July), and one assessment in the late season (8 August). In total, 100 fruit per tree (with a minimum of 25 per tree if the tree was lightly cropped) were examined visually for each assessment. A lower number for minimum number of fruit per tree was selected (25 fruit versus 50 fruit in the Geneva assessments) due to several of the trees being very lightly cropped with less available replication in terms of the number of trees—we felt the inclusion of such data, was preferable to simply eliminating a particular accession from the statistical analysis).

For all field assessments, mean percentages of fruit damaged by plum curculio were analyzed using a one-way analysis of variance (ANOVA) of arcsine-transformed data ( $P < 0.05$ ). Means were separated using Fisher protected least significant difference (LSD) (SAS Institute 2002). Names and locations of all trees examined at both Geneva and Kearneysville

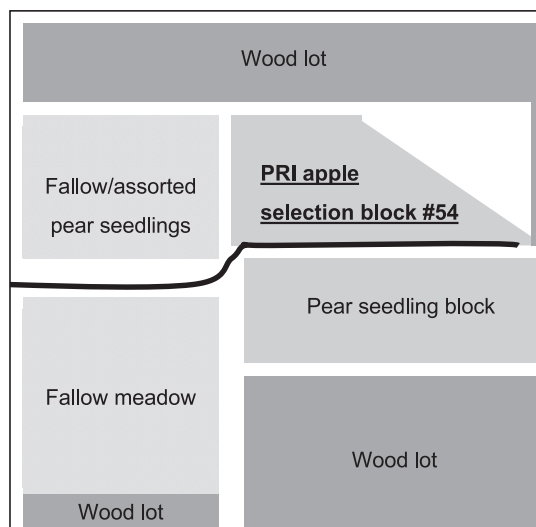


Fig. 2. Layout map of block #54 housing PRI *Malus* selections, and the surrounding landscape, Kearneysville, WV, 2005–2006. Map not exactly to scale.

are listed with their corresponding USDA germplasm resources information network (USDA 2006; [www.ars-grin.gov](http://www.ars-grin.gov)) identification numbers (i.e., PI numbers) in Table 1.

**Plum Curculio Rearing.** Adult plum curculios used in all laboratory experiments were derived from a colony housed and maintained at the USDA–AFRS. Adults were  $\approx 3$ –4 wk old and reared in the laboratory at 25°C and a photoperiod of 14:10 (L:D) h on a diet of green thinning apples based on the methods of Amis and Snow (1985). Before testing, adults were sexed by abdominal morphology according to the methods of Thomson (1932), held in groups of  $\approx 40$  males or females in wax-coated cups (473 ml) with clear plastic lids, and then they were starved for 24 h. Although they were deprived of food for this period, they were provided with a source of water (cotton dental wick saturated with distilled water).

**Adult Feeding and Oviposition Assays and Larval Survival, 2006.** Both no-choice and single-choice adult feeding and oviposition assays were conducted in 2006. Fruit for all laboratory studies was collected from the USDA–PGRU core *Malus* germplasm collection described previously. Fruit were picked and shipped to Kearneysville, where laboratory assays were conducted. Fruit were kept in a cold storage room ( $\approx 5 \pm 1^\circ\text{C}$ ) until they were used in a given bioassay. For no-choice assays, single plum curculio adults (as described previously) were placed with a single fruit and  $\approx 3$ -cm segment of wet cotton dental wick (Absorbal, Wheat Ridge, CO) saturated with distilled water inside an  $\approx 30$ -ml (1-oz.) clear plastic shot cup (JetWare, Jet Plastica Industries, Hatfield, PA) capped with a translucent plastic lid (Dixie, GA-Pacific Corp., Atlanta, GA) for 72 h. Each lid was pierced several times with a pin to provide holes for ventilation and to prevent excessive condensation. For

both males and females, there were 10 replicates each of 13 different *Malus* accessions or selections, including control treatments of 'Delicious,' 'Liberty,' and insecticide-treated Liberty. For this treatment, Liberty apples were dipped in a solution of phosmet (Imidan 70W insecticide, Gowan Co. LLC, Yuma, AZ) equivalent to the label dilute field rate (1.19 g formulated product/liter) for apples.

All adults on fruit were placed in a temperature controlled walk-in growth chamber (Convion, Controlled Environments, Ltd., Winnipeg, MB, Canada) set at  $25 \pm 2^\circ\text{C}$  and a photoperiod of 16:8 (L:D) h. Water wicks in each cup were replenished daily. Cups were checked daily for adult survival, and damage to fruit was recorded daily. After 72 h, curculio adults were removed from fruit and returned to the colony. The number of scars per fruit was analyzed using a one-way ANOVA of natural log (ln)-transformed data ( $P < 0.05$ ) (SAS Institute 2002). Independent analyses were conducted for male feeding damage, female feeding damage, and oviposition injury. Means were separated using Fisher protected LSD test (SAS Institute 2002).

After final examination of fruit for damage, all fruit exposed to females were pooled by treatment and kept in  $\approx 470$ -ml plastic cups provisioned with a moist paper towel. These fruit were kept in the same growth chamber as described above, and they were observed for 14–20 d for emergence of larvae from the eggs deposited in fruit by females. Larval survival rates were calculated for each treatment by dividing the number of emerged larvae by the total number of oviposition scars on the group of fruit. Pairwise comparisons of larval survival proportions between treatments were made using a Z test ( $P < 0.05$ ) (Minitab, Inc. 2005).

For choice assays, all methodology was the same as described above except that a single untreated Delicious fruit was added to each cup to provide a single choice between two fruit for each adult. The Delicious control fruit were marked with a small  $<1$ -mm speck by using a felt tipped colored pen. For the Delicious treatment, the marked Delicious control was paired with an unmarked Delicious fruit to ensure there was no attractant or deterrent effect of the marking. Ten replicates of each treatment were examined for 72 h, and daily counts were made of all damage done to both fruits in each cup. For each treatment, feeding damage, oviposition scars, or both were compared between Delicious control fruit versus experimental fruit by using a paired *t*-test analysis ( $P < 0.05$ ) of ln-transformed data. A significant *t* value indicated a significant preference for one fruit over the other, whereas a nonsignificant *t* value indicated no preference. Larval survival from fruit used in choice assays was not assessed.

No-choice and choice assays were each conducted twice over the 2006 growing season. All fruit for the first set of assays was harvested in Geneva, NY, on 7 June and immediately shipped to Kearneysville. The first no-choice assay was initiated on 13 June, and the first choice assay was initiated on 27 June. All fruit for the second set of assays was harvested in Geneva, NY,

**Table 2.** Mean  $\pm$  SEM percentage of fruit damaged by plum curculio in the field on various *Malus* accessions housed at the USDA Plant Genetic Resources Unit (PGRU) core collection in Geneva, NY, during a mid-season (28 June) assessment in 2005

| Accession                       | <i>n</i> <sup>a</sup> | % $\pm$ SEM <sup>b</sup> |
|---------------------------------|-----------------------|--------------------------|
| <i>M. × domestica</i> Delicious | 2                     | 3.34 (3.34)abcd          |
| <i>M. × domestica</i> Liberty   | 4                     | 19.17 (10.24)d           |
| <i>M. × domestica</i> Emilia    | 3                     | 0.00 (0.00)a             |
| <i>M. hybrid</i> PRI 1312-6     | 3                     | 7.30 (3.51)abcd          |
| <i>M. hybrid</i> PRI 1732-6     | 2                     | 1.43 (1.43)abc           |
| <i>M. hybrid</i> Co-op 15       | 2                     | 14.52 (11.19)cd          |
| <i>M. hybrid</i> E14-32         | 1                     | 10.00 (0.00)abcd         |
| <i>M. hybrid</i> E29-56         | 3                     | 10.95 (3.72)abcd         |
| <i>M. hybrid</i> E36-7          | 3                     | 2.86 (1.65)abc           |
| <i>M. hybrid</i> E7-47          | 2                     | 5.00 (0.71)abcd          |
| <i>M. hybrid</i> E7-54          | 2                     | 11.43 (4.29)bcd          |
| <i>M. hybrid</i> E31-10         | 3                     | 14.55 (12.77)cd          |
| <i>M. hybrid</i> E11-24         | 2                     | 14.29 (0.00)cd           |
| <i>M. gunnanensis</i> Vilmorin  | 3                     | 0.48 (0.48)ab            |
| <i>M. bhutanica</i> Macrocarpa  | 3                     | 2.38 (1.72)abc           |
| <i>M. fusca</i>                 | 3                     | 1.43 (0.83)abc           |
| <i>M. × robusta</i> Korea       | 4                     | 0.72 (0.41)ab            |
| <i>M. sieboldii</i>             | 3                     | 0.00 (0.00)a             |

<sup>a</sup> Number of trees examined.

<sup>b</sup> Means followed by the same letter are not significantly different ( $P < 0.05$ ; Fishers's LSD, by using arcsine-transformed data).

on 22 June, with the second no-choice assay initiated on 4 July and the second choice assay initiated on 19 July. Fruit size varied considerably among the different *Malus* accessions, and it likely had an effect on the observed preferences for feeding and oviposition and on larval survival rates due to limitations on the amount of fruit surface area and fruit flesh volume.

## Results

Plum curculio response to various apple and crabapple fruit was quite variable over the course of 2 yr of field and laboratory assessments, with plum curculio ultimately able to inflict feeding or oviposition damage on all accessions. An early season field assessment conducted in 2005 indicated significant differences ( $F = 2.16$ ;  $df = 17, 30$ ;  $P = 0.0319$ ) among various *Malus* accessions housed in the USDA-PGRU core collection. Fruit of *M. × domestica* 'Emilia' and the crabapple species *M. sieboldii* (Regel) Rehd. were free of any plum curculio damage, and a total of eight *Malus* accessions exhibited damage levels significantly less than those found on Liberty (Table 2). This included five crabapple accessions [*M. gunnanensis* (French) Schneid., *M. fusca* (Raf.) Schneid., *M. bhutanica* (W. W. Sm.) J. B. Phipps, *M. × robusta* (Carr.) Rehd., and *M. sieboldii*]. Two of the *Malus* releases previously described as resistant to plum curculio (PRI 1732-2 and E36-7) exhibited levels of damage significantly lower than Liberty. Although these two cultivars had less damage than Liberty, they did not have significantly less damage than several other accessions (Table 2).

Despite tremendous variation in numerical damage values, there were no significant differences in the percentage of plum curculio fruit damage in 2006 during either mid-season ( $F = 0.75$ ;  $df = 14, 34$ ;  $P =$



**Table 3.** Mean  $\pm$  SEM percentage of fruit damaged by plum curculio in the field for various *Malus* accessions housed at the USDA Plant Genetic Resources Unit (PGRU) core collection in Geneva, NY, during mid-season (20 June) and late season (9 Aug.) assessments in 2006

| Accession                       | n | 2006 % damage, mid-season ( $\pm$ SEM) | n | 2006 % damage, late season ( $\pm$ SEM) |
|---------------------------------|---|--|---|---|
| <i>M. × domestica</i> Delicious | 3 | 11.33 (7.31)                           | 3 | 53.00 (8.02)                            |
| <i>M. × domestica</i> Liberty   | 3 | 3.33 (0.67)                            | 4 | 48.50 (6.83)                            |
| <i>M. × domestica</i> Emilia    | * |  | * |   |
| <i>M. hybrid</i> PRI 1312-6     | 4 | 2.67 (1.04)                            | 4 | 68.50 (2.33)                            |
| <i>M. hybrid</i> PRI 1732-6     | 4 | 6.00 (3.97)                            | 4 | 51.25 (6.56)                            |
| <i>M. hybrid</i> Co-op 15       | 3 | 9.33 (6.36)                            | 3 | 51.33 (15.51)                           |
| <i>M. hybrid</i> E14-32         | 2 | 4.50 (1.50)                            | 3 | 50.67 (5.33)                            |
| <i>M. hybrid</i> E29-56         | 4 | 7.67 (2.75)                            | 4 | 79.75 (7.00)                            |
| <i>M. hybrid</i> E36-7          | 2 | 0.50 (0.50)                            | 2 | 50.00 (12.00)                           |
| <i>M. hybrid</i> E7-47          | 4 | 2.33 (1.26)                            | 3 | 49.00 (12.42)                           |
| <i>M. hybrid</i> E7-54          | 3 | 3.00 (2.08)                            | 2 | 58.50 (9.50)                            |
| <i>M. hybrid</i> E31-10         | 4 | 3.33 (1.61)                            | 3 | 65.67 (6.44)                            |
| <i>M. hybrid</i> E11-24         | 2 | 7.50 (7.50)                            | 2 | 70.00 (8.00)                            |
| <i>M. bhutanica</i> Macrocarpa  | 3 | 1.33 (0.67)                            | 2 | 48.50 (18.50)                           |
| <i>M. × robusta</i> Korea       | 4 | 4.33 (1.61)                            | * |   |
| <i>M. sieboldii</i>             | 4 | 1.00 (0.50)                            | 3 | 23.33 (8.45)                            |

Asterisk (\*) indicates that field data was unavailable or not collected.

0.712) or late season ( $F = 2.04$ ;  $df = 13, 41$ ;  $P = 0.055$ ) assessments (Table 3). Overall levels of early season damage on 28 June 2005 were slightly higher ( $<19\%$ ) than on 20 June 2006 ( $<12\%$ ), with more damage (from 23 to 79%) occurring by 9 August 2006. Similarly, there were no significant differences observed in early season ( $F = 2.32$ ;  $df = 6, 12$ ;  $P = 0.165$ ), mid-season ( $F = 1.03$ ;  $df = 6, 16$ ;  $P = 0.461$ ) or late season ( $F = 1.23$ ;  $df = 6, 12$ ;  $P = 0.414$ ) fruit damage from field assessments in a separate planting of *Malus* hybrid selections in Kearneysville (Table 4). Plum curculio damage levels were very high ( $>25\%$ ) in all accessions assessed at this Kearneysville site, including one accession (E36-7) that has been classified as “resistant” to plum curculio (Goonewardene 1987).

In an early season no-choice assay with fruit harvested 7 June 2006, there were significant differences in the level of feeding damage done by both male ( $F = 5.78$ ;  $df = 11, 108$ ;  $P < 0.0001$ ) and female ( $F = 6.72$ ;  $df = 11, 108$ ;  $P < 0.0001$ ) plum curculio adults. Female oviposition damage also varied significantly ( $F = 6.99$ ;  $df = 11, 108$ ;  $P < 0.0001$ ). Similarly, in a mid-season assay using fruit harvested on 22 June 2006, female feeding ( $F = 4.17$ ;  $df = 11, 108$ ;  $P < 0.0001$ ), male feeding ( $F = 3.36$ ;  $df = 11, 105$ ;  $P = 0.0005$ ), and female oviposition ( $F = 3.46$ ;  $df = 11, 108$ ;  $P = 0.0004$ ) damage

varied significantly among the observed accessions (Table 5). Early season feeding damage by females was greatest on *M. bhutanica* ‘Macrocarpa’ and *M. × robusta* ‘Korea’ and males fed on Emilia, Korea, and *M. sieboldii* more than other accessions. Later in the season, feeding by females was greatest on Emilia and PRI 1312-6, whereas males fed more on E36-7 than Korea and *M. sieboldii*, which had more feeding than all other accessions (Table 5). However, with female oviposition damage during the early season assay, there were four accessions (*M. fusca*, *M. × robusta*, *M. sieboldii*, and Co-op 15) that had significantly less damage than Liberty, Emilia, and Delicious, whereas later in the season, oviposition was higher on Macrocarpa and Liberty than most other accessions (Table 5).

Adults of both sexes exhibited a significant preference in feeding, and females exhibited a significant preference in oviposition for the Delicious control compared with four accessions (*M. yunnanensis*, *M. bhutanica*, *M. fusca*, *M. × robusta*) during an early season choice assay (Table 6). Females also exhibited a significant preference for feeding on the Delicious compared with fruit of Co-op 15, but there was no difference in oviposition preference. Males exhibited a significant feeding preference for the Delicious control compared with PRI 1732-2, but there was no sig-

**Table 4.** Mean  $\pm$  SEM percentage of fruit damaged by plum curculio in the field for various *Malus* selections with claimed “pest resistance” housed at the Appalachian Fruit Research Station, block #54 in Kearneysville, WV, during an early season (8 June), mid-season (13 July), and late season (8 Aug.) assessments in 2006

| Accession               | n | 2006 % damage, early season ( $\pm$ SEM) | n | 2006 % damage, mid-season ( $\pm$ SEM) | n | 2006 % damage, late season ( $\pm$ SEM) |
|-------------------------|---|--|---|--|---|---|
| <i>M. hybrid</i> E14-32 | 2 | 25.00 (0.00)                             | 4 | 59.26 (8.53)                           | 4 | 36.45 (15.54)                           |
| <i>M. hybrid</i> E29-56 | 2 | 41.54 (9.46)                             | 2 | 43.50 (7.50)                           | 2 | 53.62 (9.62)                            |
| <i>M. hybrid</i> E36-7  | 1 | 25.00 (0.00)                             | 1 | 30.00 (0.00)                           | * |   |
| <i>M. hybrid</i> E7-47  | 1 | 47.00 (0.00)                             | 1 | 27.00 (0.00)                           | 1 | 27.00 (0.00)                            |
| <i>M. hybrid</i> E7-54  | 3 | 28.95 (2.64)                             | 4 | 51.77 (8.41)                           | 1 | 71.70 (0.00)                            |
| <i>M. hybrid</i> E31-10 | 2 | 36.89 (10.89)                            | 3 | 55.75 (10.40)                          | 1 | 92.00 (0.00)                            |
| <i>M. hybrid</i> E11-24 | 2 | 54.25 (8.20)                             | 2 | 56.50 (3.50)                           | 2 | 50.00 (0.00)                            |

Asterisk (\*) indicates that field data was unavailable due to lack of fruit.

**Table 5.** Average fruit diameter estimates (millimeters) and mean  $\pm$  SEM number of female feeding and oviposition scars and male feeding scars on fruit collected during the early season (7 June) and mid-season (22 June) after 72-h exposure to single fruit in a no-choice assay with various *Malus* accessions in the laboratory, 2006

| Accession                      | Mean $\pm$ SEM damage on fruit from the early season <sup>a</sup> |               |                |               | Mean $\pm$ SEM damage on fruit from the mid-season <sup>a</sup> |               |                |               |
|--------------------------------|---|---------------|----------------|---------------|---|---------------|----------------|---------------|
|                                | Avg. fruit diam.  | ♀ Feeding     | ♀ Oviposition  | ♂ Feeding     | Avg. fruit diam.  | ♀ Feeding     | ♀ Oviposition  | ♂ Feeding     |
| Liberty dipped in Imidan       | 18  | 0.5 (0.17)a   | 0.0 (0.00)a    | 0.3 (0.15)a   | 35  | 0.3 (0.21)a   | 0.1 (0.10)a    | 0.0 (0.00)a   |
| Liberty                        | 18  | 6.7 (2.17)bc  | 17.5 (4.17)f   | 3.0 (0.71)cde | 35  | 8.1 (1.23)bc  | 9.3 (2.71)de   | 0.5 (0.31)abc |
| Delicious                      | 17  | 4.8 (1.06)b   | 11.8 (1.91)ef  | 2.9 (1.13)bcd | 34  | 6.3 (1.56)bc  | 6.0 (2.31)bcde | 0.6 (0.36)abc |
| Emilia                         | 14  | 3.5 (0.60)b   | 16.3 (3.73)f   | 4.8 (1.13)def | 26  | 12.1 (2.70)c  | 7.0 (1.75)cde  | 1.2 (0.42)bc  |
| <i>M. yunnanensis</i> Vilmorin | 6   | 5.0 (1.30)b   | 8.5 (2.80)cdef | 1.9 (0.69)bc  | 10  | 6.9 (1.39)bc  | 2.3 (1.26)ab   | 1.6 (0.65)bc  |
| <i>M. bhutanica</i> Macrocarpa | 9   | 14.8 (3.06)e  | 10.9 (2.59)def | 1.9 (0.62)bc  | 14  | 5.0 (1.67)b   | 12.8 (4.03)e   | 0.2 (0.20)ab  |
| <i>M. fusca</i>                | 4   | 7.4 (1.53)bcd | 2.0 (1.12)ab   | 2.6 (1.01)bc  |   |               |                |               |
| <i>M. × robusta</i> Korea      | 6   | 12.8 (2.78)de | 5.6 (2.08)cd   | 6.7 (1.97)ef  | 10  | 6.3 (1.58)bc  | 2.2 (1.04)ab   | 2.1 (0.84)c   |
| <i>M. sieboldii</i>            | 4   | 6.4 (1.54)bcd | 4.8 (1.19)cd   | 8.5 (1.28)f   | 6   | 8.1 (1.59)bc  | 7.7 (2.37)cde  | 2.5 (1.40)c   |
| PRI 1312-6                     | 17  | 9.6 (1.50)bcd | 9.4 (2.26)cdef | 4.6 (1.19)def | 36  | 13.7 (3.76)c  | 4.8 (0.84)cde  | 0.8 (0.42)abc |
| PRI 1732-2                     | 17  | 5.6 (1.25)ab  | 6.2 (2.06)cde  | 1.3 (0.60)ab  | 35  | 8.2 (2.24)bc  | 3.8 (1.08)bcd  | 1.2 (0.57)abc |
| Co-op 15                       | 16  | 5.7 (1.41)ab  | 3.2 (1.04)bc   | 1.9 (0.53)bc  | 30  | 7.7 (2.17)bc  | 8.0 (3.29)bcde | 1.1 (0.50)abc |
| E36-7                          |   |               |                |               | 35  | 11.7 (3.47)bc | 7.1 (2.16)cde  | 4.9 (1.35)d   |

<sup>a</sup> Means in each column followed by the same letter are not significantly different ( $P < 0.05$ ; Fisher's LSD, by using ln-transformed data).

nificant preference in either feeding or oviposition by females (Table 6). In a subsequent repetition (mid-season evaluation) of the same assay with fruit harvested later in June, however, plum curculio exhibited a preference for oviposition and male feeding for several experimental accessions over a Delicious control (Table 6).

There were numerous significant differences among accessions in the level of larval survival in fruit (Table 7). This was true for both early and mid-season assays, with *M. yunnanensis* and *M. sieboldii* exhibiting the lowest numerical levels of survival in both assays. Larval survival was  $>10\%$  among cultivars previously classified in the literature as "resistant" (PRI 1312-6, PRI 1732-2, and Co-op 15) during the early season assay and was  $>20\%$  for E36-7 and the same aforementioned cultivars during the mid-season assay.

## Discussion

Although all 206 accessions in the PGRU core collection were assessed in the field in 2005, only a sam-

pling of that data from a limited number of accessions is presented here. The accessions presented in this article (besides common commercial cultivars of Liberty and Delicious, which served as controls) were chosen for one of two reasons. Either there was a prior published claim of some type of pest resistance, or they seemed to be less susceptible to curculio attack based on our observations in 2005. E36-7 (Goonewardene 1987), PRI 1312-6, PRI 1732-2, and Co-op 15 (USDA 2006) were reported to be resistant to plum curculio. 'E14-32', 'E29-56', 'E7-47', 'E7-54', 'E31-10', and 'E11-24', although not specifically reported to be resistant to plum curculio, are reported to be resistant to other apple pests (Goonewardene 1987, Goonewardene and Howard 1989). *M. × domestica* Emilia and various exotic crabapple accessions (*M. yunnanensis*, *M. bhutanica*, *M. fusca*, *M. × robusta*, and *M. sieboldii*) stood out in 2005 as having much lower levels of plum curculio damage than the rest of the core collection. On this basis, these accessions were chosen for further study in the laboratory in 2006.

**Table 6.** Early season (fruit picked 7 June) and mid-season (fruit picked 22 June) adult feeding and oviposition preferences after 72 h of exposure, given a single choice between a fruit from a given *Malus* accession and a Delicious fruit control in the laboratory, 2006

| Accession                      | Early season preference <sup>a</sup> |               |           | Mid-season preference <sup>a</sup> |               |           |
|--------------------------------|--------------------------------------|---------------|-----------|------------------------------------|---------------|-----------|
|                                | ♀ Feeding                            | ♀ Oviposition | ♂ Feeding | ♀ Feeding                          | ♀ Oviposition | ♂ Feeding |
| Delicious                      | N                                    | N             | N         | A                                  | N             | N         |
| Liberty                        | N                                    | N             | N         | N                                  | N             | N         |
| Emilia                         | N                                    | N             | N         | A                                  | N             | N         |
| <i>M. yunnanensis</i> Vilmorin | C                                    | C             | C         | N                                  | N             | A         |
| <i>M. bhutanica</i> Macrocarpa | C                                    | C             | C         | N                                  | A             | N         |
| <i>M. fusca</i>                | C                                    | C             | C         |                                    |               |           |
| <i>M. × robusta</i> Korea      | C                                    | C             | C         | N                                  | N             | N         |
| <i>M. sieboldii</i>            |                                      |               |           | N                                  | N             | N         |
| PRI 1312-6                     | N                                    | N             | N         | A                                  | A             | A         |
| PRI 1732-2                     | N                                    | N             | C         | N                                  | A             | A         |
| Co-op 15                       | C                                    | N             | N         | A                                  | A             | A         |
| E36-7                          |                                      |               |           | N                                  | A             | A         |

<sup>a</sup> N indicates no significant preference for either fruit, A indicates a significant preference for the experimental fruit, and C indicates a significant preference for the fruit from a Delicious control fruit, as determined by a paired *t*-test ( $P < 0.05$  with ln-transformed data).

**Table 7.** Total number of oviposition scars (N) on fruit from various *Malus* accessions and the percentage of larval emergence from these fruit from early season and mid-season assays in 2006

| Accession                       | Early season |                                 | Accession                       | Mid-season |                                 |
|---------------------------------|--------------|---------------------------------|---------------------------------|------------|---------------------------------|
|                                 | N            | % larval emergence <sup>a</sup> |                                 | N          | % larval emergence <sup>a</sup> |
| <i>M. gunnanensis</i> Vilmorin  | 85           | 0.00a                           | <i>M. sieboldii</i>             | 77         | 0.00a                           |
| <i>M. sieboldii</i>             | 48           | 0.00a                           | <i>M. gunnanensis</i> Vilmorin  | 23         | 4.35ab                          |
| <i>M. fusca</i>                 | 20           | 0.00a                           | <i>M. bhutanica</i> Macrocarpa  | 128        | 12.50bc                         |
| <i>M. bhutanica</i> Macrocarpa  | 109          | 6.42b                           | <i>M. hybrid</i> PRI 1312-6     | 48         | 22.92bcd                        |
| <i>M. × robusta</i> Korea       | 56           | 7.14bc                          | <i>M. hybrid</i> E36-7          | 71         | 25.35d                          |
| <i>M. × domestica</i> Emilia    | 173          | 7.51bc                          | <i>M. × domestica</i> Liberty   | 93         | 29.03de                         |
| <i>M. × domestica</i> Delicious | 118          | 11.02bcd                        | <i>M. hybrid</i> Co-op 15       | 80         | 32.50de                         |
| <i>M. hybrid</i> PRI 1732-2     | 62           | 12.90bcd                        | <i>M. × domestica</i> Delicious | 60         | 35.00de                         |
| <i>M. hybrid</i> PRI 1312-6     | 94           | 15.96cd                         | <i>M. × domestica</i> Emilia    | 70         | 40.00de                         |
| <i>M. × domestica</i> Liberty   | 176          | 17.05d                          | <i>M. × robusta</i> Korea       | 22         | 40.91de                         |
| <i>M. hybrid</i> Co-op 15       | 32           | 25.00d                          | <i>M. hybrid</i> PRI 1732-2     | 38         | 44.74e                          |

<sup>a</sup> Pooled larval emergence values followed by a different letter are significantly different according to Z test for overlap of confidence intervals ( $P < 0.05$ ).

Further field assessments in 2006 shed new light on our initial conclusions from 2005, because observations from later in the growing season (and from a block now 2 yr removed from insecticide treatments) indicated a high level of damage was present on all accessions by the late season field assessment. Meanwhile, the Kearneysville site has been observed to have a very high resident population of plum curculio, and this is reflected in the very high levels of plum curculio damage on all accessions observed in 2006.

Laboratory assays of adult feeding and oviposition on fruit provided an even more rigorous test of possible resistance traits in these various accessions. Although significant differences were observed, all accessions exhibited some significant level of damage from either adult feeding, oviposition, or both. Indeed, no accessions were free from damage except for fruit treated with a conventional insecticide. Furthermore, choice assays revealed that plum curculio adults did not exhibit a distinct avoidance of any particular accession in a repeatable manner. Although some of the crabapple accessions seemed to be less preferred than a Delicious control in the early season assay, this was likely due to differences in relative fruit size (i.e., there was more room to feed and oviposit on the Delicious control than on the smaller crabapple alternative) than any genetically inherent resistance factors. Moreover, Alm and Hall (1986) found a significant correlation between fruit size and percentage of oviposition injury in a field survey of *Malus* spp., including one crabapple accession surveyed here, *M. sieboldii*. In the later mid-season assay, when crabapple fruit were somewhat larger, no preference for Delicious was observed. Finally, plum curculio larvae were able to feed and develop in all fruit except the smallest crabapple accessions (including early season samples of *M. fusca* and *M. gunnanensis* Vilmorin and both early and mid-season samples of *M. sieboldii*), where the mass of fruit (<5 g) was very likely the limiting factor on larval survival. Green thinning apples used to rear plum curculio in our laboratory weigh ≈20–30 g (T.C.L., unpublished data), and Amis and Snow (1985) reported using ≈12–15 g of apples, thus

much greater in weight than the accessions given above. Our observations from these studies suggest that although plum curculio larvae were able to feed successfully on even the smallest fruit, they simply did not have an adequate volume of food to complete development and emerge successfully and pupate.

Clearly, there is significant variability among *Malus* germplasm in the level of susceptibility to attack from plum curculio. However, although some accessions had levels of feeding and/or oviposition damage that was less than common commercial cultivars (such as Liberty or Delicious), all accessions exhibited economically damaging levels of plum curculio injury. Given the totality of data—from field assessments, laboratory observations of adults, and larval survival in fruit—we conclude that fruit from all observed accessions is at least somewhat suitable for plum curculio. Thus, no accessions evaluated in this study should be considered “resistant” by any standard of commercial relevance. This includes the four accessions previously described in the literature or USDA–GRIN as being resistant (Goonewardene 1987, USDA 2006). Such discrepancies need to be corrected and clarified in the horticultural literature and also in USDA’s online data resources. Furthermore, other accessions with such annotations, including those housed outside the USDA–PGRU core collection, should be assessed to clarify their true pest resistance status. Such clarifications are needed to prevent wasted efforts and future resources by breeders seeking to use confirmed pest-resistant germplasm.

Plum curculio has a broad host range, feeding on plants belonging to the Rosaceae (Maier 1990, Brown 2005), Ericaceae (Beckwith 1943; Mampe and Neunzig 1967; Jenkins et al. 2006) and Vitaceae (Jenkins et al. 2006) families; therefore, it is an oligophagous herbivore based on this multiple-family host use pattern (Bernays and Chapman 1994). Cultivated hosts in the family Rosaceae include apple; pear, *Pyrus communis* L.; peach, *Prunus persica* Batsch; apricot; sour cherry, *P. cerasus* L.; sweet cherry, *P. avium* (L.); European plum, *P. domestica* L.; and Japanese plum, *P. salicina* Lindl. Recent data indicate that Japanese plum is the

most highly preferred host, followed by European plum, peach, sweet cherry, tart cherry, apricot, apple, and pear, respectively (Leskey and Wright 2007). That plum curculio is such a significant pest of apple clearly indicates that the species is able to survive and even thrive on a less preferred host. Given the polyphagous nature of plum curculio, combined with our findings, we feel that the likelihood of finding a true source of genetic resistance within *Malus* is very low.

Although host plant resistance to plum curculio seems to be unlikely, there is reason for optimism that genetic resistance may exist in *Malus* for pest species with a more host-specific ecology. Preliminary data indicate that the USDA-PGRU core collection houses accessions that are resistant to feeding by oriental fruit moth, *Grapholita molesta* (Busck); codling moth, *Cydia pomonella* L.; and apple maggot (C.T.M., unpublished data). We hypothesize that species feeding on a more narrow host range and encountering a smaller variety of plant defenses are perhaps more likely to be susceptible to host plant resistance mechanisms expressed within a given plant genus, such as *Malus*, for example. Future studies should investigate the potential for *Malus* host resistance among some of the more monophagous pest species, limited oligophagous pest species, or both, including those species mentioned above.

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